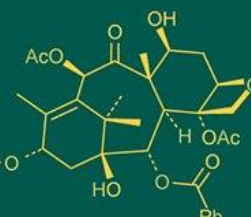
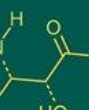
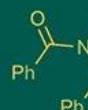


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In vitro evaluation of fungicides against *Fusarium oxysporum* f. sp. *ciceri* causing wilt of chickpea (*Cicer arietinum* L.)

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Abstract

Chickpea (*Cicer arietinum* L.) is one of the most important pulse crops worldwide; however, its productivity is severely constrained by Fusarium wilt caused by *Fusarium oxysporum* f. sp. *ciceri*. The present study was undertaken to evaluate the efficacy of eight fungicides against the pathogen under *in vitro* conditions using the poisoned food technique. The fungicides were tested at three concentrations (50, 100, and 150 ppm), and radial mycelial growth was recorded at 48, 96, and 144 hours after inoculation. Significant differences were observed among the fungicidal treatments in suppressing fungal growth. Thiophanate methyl 70% WP and Carbendazim 12% + Mancozeb 63% WP completely inhibited the mycelial growth of *F. oxysporum* f. sp. *ciceri* (100% inhibition) at all concentrations throughout the observation period. Propiconazole 25% EC also exhibited high antifungal activity, recording 86.67% and 90.00% inhibition at 100 and 150 ppm, respectively, after 144 hours. Thiram 70% WP showed 83.33% and 85.00% inhibition at 100 and 150 ppm, whereas Chlorothalonil and Mancozeb provided moderate suppression of the pathogen. Thifluzamide 24% SC was the least effective treatment, with only 68.33-70.00% inhibition at the final observation. The study demonstrates that Thiophanate methyl and Carbendazim + Mancozeb are highly effective against *F. oxysporum* f. sp. *ciceri* under laboratory conditions and may serve as promising fungicidal options for the integrated management of chickpea wilt.

Keywords: Chickpea, *Fusarium oxysporum* f. sp. *Cicero*, Fungicides, *in vitro* evaluation, poisoned food technique, Mycelial growth inhibition, wilt management

Introduction

Pulses are important source of dietary protein, and have unique property of maintaining and restoring soil fertility through biological nitrogen fixation as well as conserving, and improving physical properties of soil by virtue of their deep root system and leaf fall. Chickpea (*Cicer arietinum* L.) (diploid, $2n = 16$) is the world's most important food legume crop after dry beans, grown throughout tropical, subtropical and temperate regions in South and West Asia, East and North Africa, southern Europe, North and South America, and Australia. Approximately 13.5 mha of chickpea is cultivated in more than 50 countries worldwide that yield nearly 13.1 mmt (FAOSTAT, 2025) [4]. Of that, 89.2% is grown in Asia and accounts for 84.5% of the world production (FAOSTAT, 2025) [4]. India ranked first in area and production, followed by Pakistan, Iran and Australia with respect to area and Australia, Myanmar with respect to production. Chickpea seeds are a major source of human food and animal feed because of their high content of lysine-rich protein (Jukanti *et al.*, 2012) [5]. Chickpea seed is very rich in protein (Hulse 1991) and considered alternate to meat. It contains 38-59% protein, 3% carbohydrate, 4.8-5.5%, ash fiber 3% oil, 0.3% phosphorus and crude lipid, it also contains amino acids and polyphenol contents with some sort of minerals (Daur *et al.*, 2008) [3]. Chickpea is helpful for lowering blood cholesterol (Pittaway *et al.*, 2008) [8]. Chickpea is relatively economical and a good source of folic acid and tocopherols (Ciftci *et al.*, 2010) [2]. Chickpea is an excellent source of folic acid and contains other water-soluble vitamins like riboflavin, pantothenic acid and pyridoxine. It is rich source of vitamin A, the most important and widely distributed carotenoid is β -carotene converted to vitamin A (Abbo *et al.*, 2005) [1].

It is consumed in various forms, viz. dal, besan, crushed or whole grain, boiled or parched, green grain. Husk is fed to horses and cattle. Chickpea is attacked by a number of seed and soil borne diseases.

In recent years there has been concern about gradual decline in per capita availability of pulses. An important reason for this miserable situation in the country is mainly due to the attack of the soil borne pathogens. According to Sarwar *et al.*, (2012) ^[9] wilt is a major restrictive factor to chickpea production. Wilt of chickpea is caused by *Fusarium oxysporum f. sp. Ciceri* (FOC). It is widely distributed throughout the country and is responsible for heavy reduction in crop yield and quality.

Keeping existing research gaps in mind *in vitro* investigations were carried out to screen eight fungicides at

three concentrations through the poisoned food technique.

Materials and Methods

The experiments were conducted at the Postgraduate Laboratory, Department of Plant Pathology, SVPUAT, Meerut. The effect of eight fungicides were tested for their efficacy against *Fusarium oxysporum f. sp. ciceri* under *in vitro* condition. Chemicals were procured from store room, in the Department of Plant Pathology of S.V.P University of Agriculture & Technology Meerut (U.P.). Bioassay test was done by using poisoned food technique as given by Schmitz in 1930. Efficacy of 8 different fungicides were evaluated against *Fusarium oxysporum f. sp. ciceri*. The common name, trade name, and concentration (in ppm) of used fungicides are given in the Table 1.

Table 1: List of fungicide used in *in vitro* evaluation against *Fusarium oxysporum f. sp. ciceri*.

S. No.	Treatments	Common Name	Trade Name (a.i.)	Concentration (in ppm)
1	T ₁	Thiifluzamide	Pulsor 24% SC	50,100, 150
2	T ₂	Propiconazole	Tilt 25% EC	50, 100, 150
3	T ₃	Hexaconazole	Hexon 5% SC	50, 100, 150
4	T ₄	Thiophanate Methyl	Mildivip 70% WP	50, 100, 150
5	T ₅	Thiram	Blitox 50% WP	50, 100, 150
6	T ₆	Mancozeb	Indofil M-45	50, 100, 150
7	T ₇	Carbendazim 12% + Mancozeb 63%	Policy WP	50, 100, 150
8	T ₈	Chlorothalonil	Kavach 78% WP	50, 100, 150
9	T ₉	Control		

The required concentrations of chemicals were prepared and incorporated into sterilized, cooled Potato Dextrose Agar. 20 ml of poisoned media was poured into 90 mm sterilized petri plates and allowed to solidify in aseptic condition. All plates were inoculated with actively growing 5 mm mycelial disc of *Fusarium oxysporum f. sp. ciceri*. Three replications were maintained for each treatment. These plates were incubated at 26±1 °C for five days in a B.O.D. Incubator and colony diameter was recorded. Percent inhibition of mycelial growth over control was calculated by using the formula given by Vincent (1947).

$$\% \text{ Inhibition of mycelial growth} = \frac{C-T}{C} \times 100$$

Where,

C = Growth of mycelium in control

T = Growth of mycelium in treatment

Observation recorded

- Radial growth of the pathogen.
- Percentage (%) growth inhibition.

An *in vitro* experiment was conducted to find out efficacy of 8 different fungicides viz., Thiifluzamide 24% SC, Propiconazole 25% EC, Hexaconazole 5% SC, Thiophanate Methyl 70% WP, Thiram 70%, Mancozeb 75% WP, Carbendazim 12% + Mancozeb 63% and Chlorothalonil 75% WP at three different concentrations of 50, 100 and 150 ppm by Poisoned food technique. The radial growth of pathogen was recorded at 48 hours, 96 hours and 144 hours respectively after inoculation. Thiophanate Methyl 70% WP and Carbendazim 12% + Mancozeb 63% exhibited complete inhibition of *Fusarium oxysporum f. sp. ciceri* at all three concentrations (50 ppm, 100ppm and 150ppm), Propiconazole 25% EC at 100ppm and 150ppm concentration (86.67% and 90% respectively) and Thiram at

100 ppm and 150ppm concentration (83.33%, 85.00% respectively) also gave promising results. The results revealed that there is significant difference in percent inhibition of mycelial growth of *Fusarium oxysporum f. sp. ciceri* with tested fungicides. Result in tabulated form have been presented in Table No 2, 3, 4 and Fig. No. 1, 2, 3 and Plate no 7 a and b The least inhibition percent of mycelial growth was recorded (68.33%, 68.33% and 70%) in Thiifluzamide 24% SC at all three concentrations (50ppm, 75ppm and 100ppm) after 144 hrs. incubation.

8 different fungicides viz., Thiifluzamide 24% SC, Propiconazole 25% EC, Hexaconazole 5% SC, Thiophanate Methyl 70% WP, Thiram 70%, Mancozeb 75% WP, Carbendazim 12% + Mancozeb 63% and Chlorothalonil 75% WP at three different concentrations of 50, 100 and 150 ppm were screened for their efficiency by Poisoned food technique. The radial growth of pathogen was recorded at 48 hours, 96 hours and 144 hours respectively after inoculation. Thiophanate Methyl 70% WP and Carbendazim 12% + Mancozeb 63% exhibited complete inhibition of *Fusarium oxysporum f. sp. ciceri* at all three concentrations i.e., 50 ppm, 100ppm and 150ppm. Propiconazole 25% EC (86.67%, 90% respectively) and Thiram (83.33%, 85.00% respectively) at 100 and 150ppm concentration also gave promising results. The least inhibition percent of mycelial growth was recorded (68.33%, 68.33% and 70%) in Thiifluzamide 24% SC at all three concentrations (50ppm, 75 ppm and 100 ppm) after 144 hrs. incubation respectively. Similarly, Mayur *et al.* (2001) ^[7] tested different fungicides against *Fusarium oxysporum f.sp. ciceri*. Among the systemic fungicides, Carbendazim was the most effective inhibiting the radial mycelial growth by 100% even at the lowest concentration and among the non-systemic fungicides thiram was most effective in inhibiting the radial and mycelia growth (90.98%) even at the lowest concentration. Magar *et al.* (2020) ^[6] tested different fungicides against

Fusarium oxysporum f. sp. *ciceri*. Among all significantly less mycelial growth was recorded with the Tebuconazole, Carboxin + Thiram and Carbendazim + Mancozeb and was

00 mm followed by Thiophanate methyl and Carbendazim (10 mm), Captan (17.66 mm) and Azoxystrobin (47.33 mm) as compared to highest growth (90 mm) in control.

Table 2: Effect of different fungicides on the radial growth of *Fusarium oxysporum* f. sp. *ciceri* @ 50 ppm *in vitro*

Treatments	Inhibition after 48 hours		Inhibition after 96 hours		Inhibition after 144 hours	
	Radial Growth of pathogen (mm)	% Inhibition	Radial Growth of pathogen (mm)	% Inhibition	Radial Growth of pathogen (mm)	% Inhibition
Thi fluzamide (T ₁)	8	60.00	17	57.50	19	68.33
Propiconazole (T ₂)	6	70.00	8	80.00	11	81.67
Hexaconazole (T ₃)	9	55.00	14	65.00	17	71.67
Thiophanate Methyl (T ₄)	0	100.00	0	100.00	0	100.00
Thiram (T ₅)	7	65.00	9	77.50	11	81.67
Mancozeb (T ₆)	9	55.00	11	72.50	13	78.33
Carbendazim 12% + Mancozeb 63% (T ₇)	0	100.00	0	100.00	0	100.00
Chlorothalonil (T ₈)	7	65.00	9	77.50	12	80.00
Control (T ₉)	20		40		60	
SE(m)	0.667		0.679		0.692	
CD (5%)	1.401		1.410		1.421	

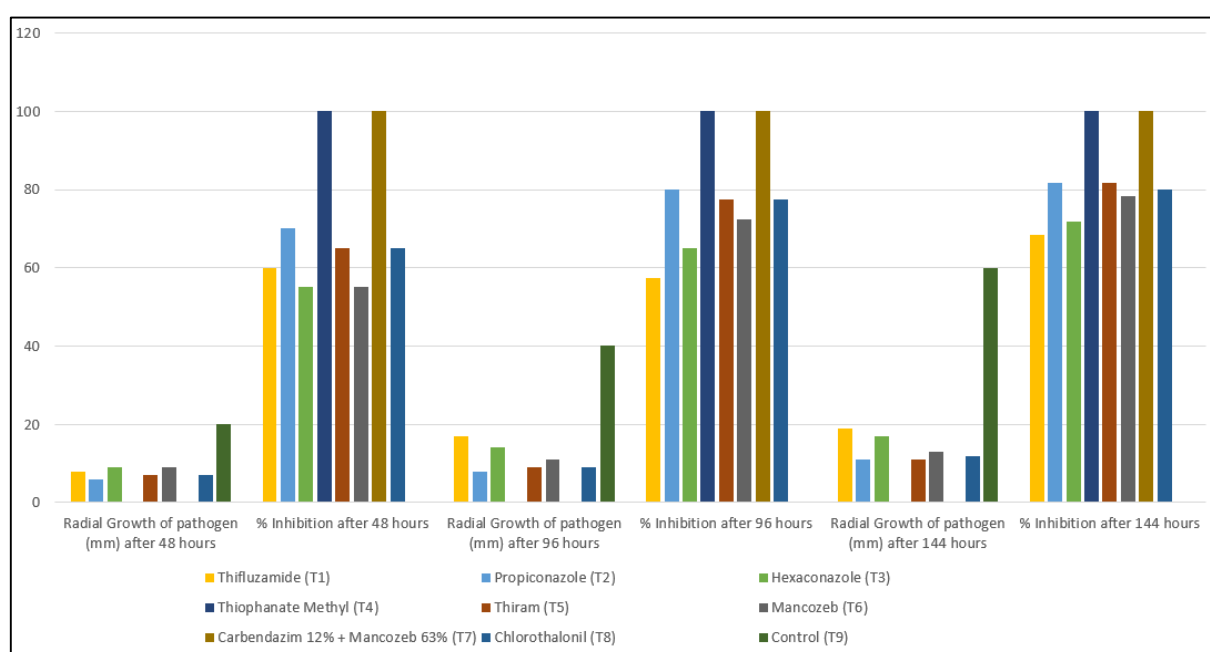


Fig 1: Effect of different fungicides on the radial growth of *Fusarium oxysporum* f. sp. *ciceri* @ 50 ppm *in vitro*

Table 3: Effect of different fungicides on the radial growth of *Fusarium oxysporum* f. sp. *ciceri* @ 100 ppm *in vitro*

Treatments	Inhibition after 48 hours		Inhibition after 96 hours		Inhibition after 144 hours	
	Radial Growth of pathogen (mm)	% Inhibition	Radial Growth of pathogen (mm)	% Inhibition	Radial Growth of pathogen (mm)	% Inhibition
Thi fluzamide (T ₁)	7	65.00	17	57.50	19	68.33
Propiconazole (T ₂)	6	70.00	7	82.5	8	86.67
Hexaconazole (T ₃)	8	60.00	13	67.50	15	75.00
Thiophanate Methyl (T ₄)	0	100.00	0	100.00	0	100.00
Thiram (T ₅)	6	70.00	9	77.50	10	83.33
Mancozeb (T ₆)	8	60.00	11	72.50	12	80.00
Carbendazim 12% + Mancozeb 63% (T ₇)	0	100.00	0	100.00	0	100.00
Chlorothalonil (T ₈)	7	65.00	9	77.50	11	81.67
Control (T ₉)	20		40		60	
SE(m)	0.667		0.692		0.833	
CD (5%)	1.401		1.410		1.624	

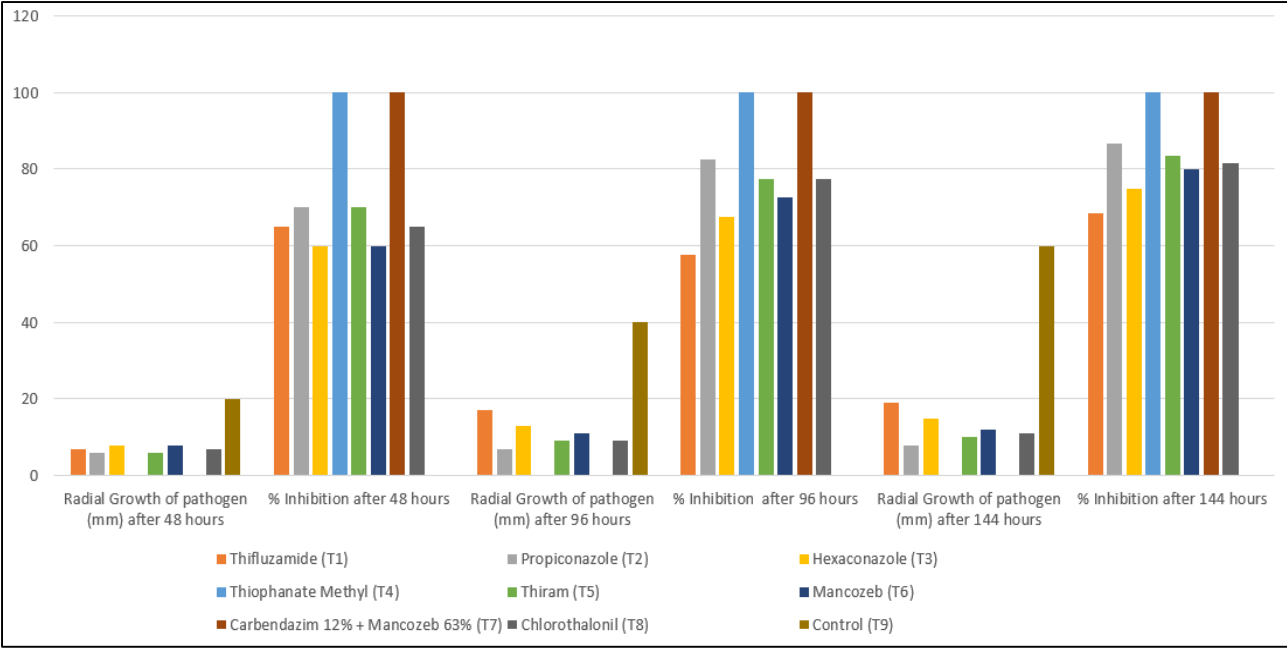


Fig 2: Effect of different fungicides on the radial growth of *Fusarium oxysporum* f. sp. *ciceri* @ 100 ppm in vitro

Table 4: Effect of different fungicides on the radial growth of *Fusarium oxysporum* f. sp. *ciceri* @ 150 ppm in vitro

Treatments	Inhibition after 48 hours		Inhibition after 96 hours		Inhibition after 144 hours	
	Radial Growth of pathogen (mm)	% Inhibition	Radial Growth of pathogen (mm)	% Inhibition	Radial Growth of pathogen (mm)	% Inhibition
Thifluzamide (T ₁)	7	65.00	16	60.00	18	70.00
Propiconazole (T ₂)	5	75.00	6	85.00	6	90.00
Hexaconazole (T ₃)	7	65.00	11	72.50	12	80.00
Thiophanate Methyl (T ₄)	0	100.00	0	100.00	0	100.00
Thiram (T ₅)	6	70.00	8	80.00	9	85.00
Mancozeb (T ₆)	7	65.00	9	77.5	11	81.67
Carbendazim 12% + Mancozeb 63% (T ₇)	0	100.00	0	100.00	0	100.00
Chlorothalonil (T ₈)	6	70.00	9	77.50	10	83.33
Control (T ₉)	20		40		60	
SE(m)	0.667		0.657		0.593	
CD (5%)	1.401		1.371		1.321	

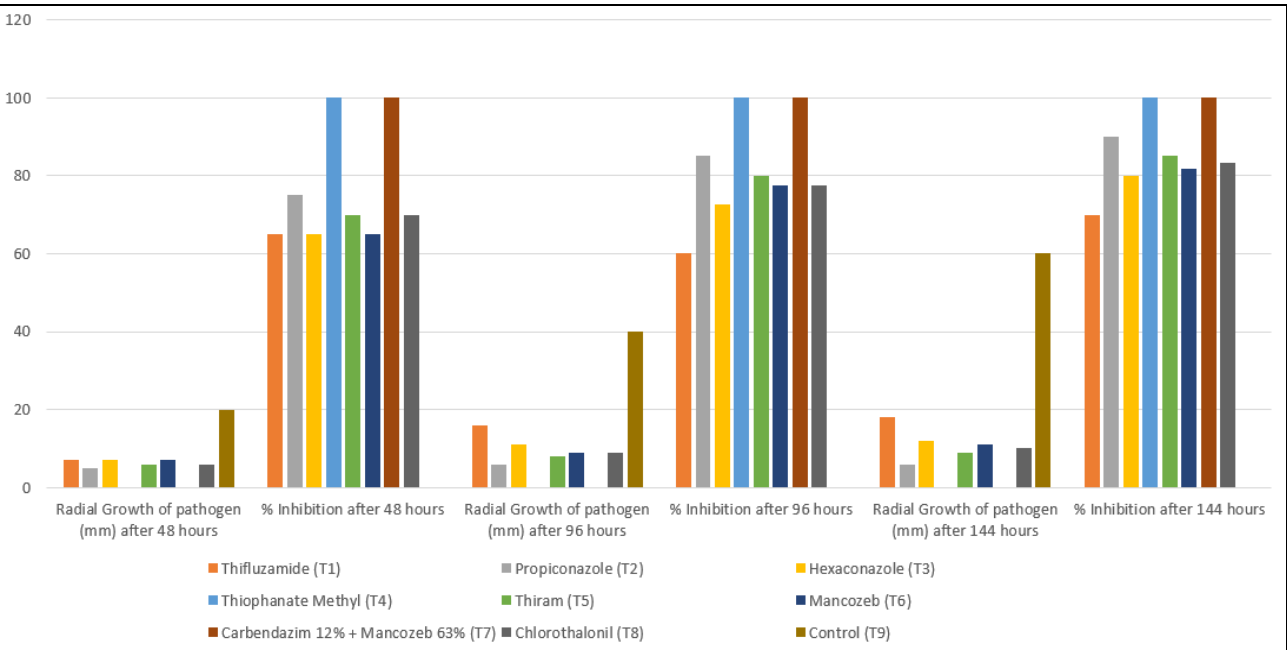


Fig 3: Effect of different fungicides on the radial growth of *Fusarium oxysporum* f. sp. *ciceri* @ 150 ppm in vitro

Conclusion

The present investigation demonstrated significant differences in the efficacy of fungicides against *Fusarium oxysporum* f. sp. *ciceri* under laboratory conditions. Thiophanate methyl 70% WP and Carbendazim 12% + Mancozeb 63% WP consistently achieved complete (100%) inhibition of mycelial growth at 50, 100, and 150 ppm throughout the incubation period, indicating their superior fungitoxic activity. Propiconazole 25% EC was the next most effective treatment, recording up to 90.00% inhibition at 150 ppm after 144 hours, followed by Thiram 70% WP (85.00%). In contrast, Thifluzamide 24% SC exhibited the lowest inhibitory effect, with only 68.33-70.00% inhibition at the final observation. These findings suggest that Thiophanate methyl and Carbendazim + Mancozeb possess excellent potential for the effective management of chickpea wilt caused by *F. oxysporum* f. sp. *ciceri*. However, further validation under greenhouse and field conditions is necessary before recommending these fungicides as part of an integrated disease management strategy.

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